STUDIES ON THE PARASITES OF THE TER-MITES II. ON *TRICHOMITUS TERMITIDIS*, A POLYMASTIGOTE FLAGELLATE WITH A HIGHLY DEVELOPED NEUROMOTOR SYSTEM

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INTRODUCTION

The occurrence in polymastigote flagellates of a structurally inte-grated fibrillar complex consisting of centrosome, rhizoplast, blepharo-plast, axostyle, undulating membrane, parabasal body, and flagella in *Trichomonas* was described by us (1915) and the complex designated as an extra-nuclear motor apparatus. Its analogy to the neuromotor apparatus of the ciliate *Diplodinium* was noted.

In the following year the more highly specialized and intimately integrated fibrillar apparatus of the binucleate diplozooic *Giardia* was definitely designated (Kofoid and Christiansen, 1916) as the neuromotor apparatus. In a paper read before the Second Pan-American Scientific Congress at Washington, January 7, 1916, the senior author extended the neuromotor conception to the flagellates generally to include the centrosome-blepharoplast and its external and internal fibrillar derivatives and connections under the name of the neuromotor apparatus.

It is the purpose of this paper to describe the neuromotor apparatus or system of one of the simpler trichomonads in which there is no axostyle but in which there occurs in response to the parasitic habit an exceptionally massive development and structural continuity of the several elements of this coördinating organ system. The organism also presents a prophetic prolongation of the period of existence of the paradesmose and of the incipient stage of mitosis,—features which are strongly suggestive of a tendency which, if continued, might well culminate in the evolution of the diplozooic flagellates, such as *Giardia*. The potency of the biochemical environment of parasites in bringing to expression latent possibilities of the organization of the living substance is once again demonstrated in this flagellate of those extraordinarily parasitized insects, the termites.

OCCURRENCE

This flagellate has been found abundantly in *Termopsis angusti-collis* Walk., a large termite commonly found in decayed oak trees on the University campus. The flagellate infests the posterior and midregions of the intestinal tract of the termite with only a slight infection or none at all of the anterior region. It is found in the lumen of the canal with no attachment to the wall. Associated with it is a large *Trichonympha*, and in cross-section of the entire intestinal tract it is found that these two flagellates, with the latter usually predominating, completely fill the lumen of the canal.

Almost every individual of this species of termite which has been examined has been found to harbor these flagellates. The number in a single host may vary greatly as it or the trichonymph may be the dominant form. In some instances the latter species may be rare with *Trichomitus termitidis* present in vast quantities. These are sporadic cases, however, with no indications of a rhythmical cycle

that is seasonal in its occurrence, as shown by examinations of the host which have been made throughout the year.

Trichomitus greatly resembles its near relative *Trichomonas* in its activities. It is, however, difficult to keep these flagellates alive in cultures, hence observations on the active forms have been limited. The extreme fragility of the cytoplasmic body as contrasted with the stout, persistent parabasal body, is particularly striking in preparations of living material. A few seconds usually suffices, in ordinary tap water, for the dissolution of the protoplasm, leaving the neuro-motor system still intact.

Nutrition in *Trichomitus* is holozoic. It, like *Trichonympha*, is evidently only a commensal, or at least is not truly parasitic, i.e., living on the tissues or fluids of the host. The food particles found within the cytoplasm consist principally of woody fibers upon which the termite feeds.

MORPHOLOGY

The morphology of this species of *Trichomitus* is of especial signifi-cance not only in view of the distinctness with which the neuromotor organ system is developed and integrated but also in the unquestion-able certainty with which the relationship of the centrosome to the blepharoplast is established, as will be shown later. The relatively large size of the organisms (75 to 150μ) and their abundance have made possible an analysis of these structures not obtainable with the smaller trichomonads of our earlier studies (Kofoid and Swezy, 1915).

SHAPE AND SIZE OF BODY

The body of *Trichomitus termitidis* is exceedingly amoeboid and protean in life, having neither constancy of form nor resistance to deformation on contact with other organisms or objects. Its periplast is unusually thin and delicate and in the larger forms especially is not infrequently ruptured in the making of smear preparations. It has nevertheless a certain characteristic range of forms within which, in free movement, it is seen or preserved on fixation. These vary from the asymmetrical pyriform contour, with the large end anterior and the posterior tapering to a blind point (pl. 3, fig. 1), to the ellipsoidal (pl. 3, fig. 14) or subspheroidal shape (pl. 3, fig. 5), with the slightly greater diameter posterior to the center.

The factors conducing to these changes in form are the stages of general contraction of the body, the mass of food vacuoles which is usually greater in the more rounded forms, and the proportional length of the parabasal body and undulating membrane. As a result of multiple fission (pl. 4, figs. 28, 30) one of the daughters receives the ancestral parabasal and two of its associated flagella, both of which are disproportionally large for the cytoplasmic mass of this schizont. Regulative resorbtion, in (pl. 4, fig. 31) or out of a cyst, or rapid cytoplasmic growth, would be necessary to readjust the volumetric relation of the neuromotor system and the cytoplasmic mass.

The range in size in this species is very considerable (fig. A, 1–7). The smallest schizonts we have recorded (pl. 4, fig. 32) are but 16μ in length while the largest exceed 200μ. These giant individuals are probably approaching multiple fission. They rarely survive the smearing operation intact. Not infrequently the nucleus and its attached neuromotor system of such giant individuals will be found intact and still active after the loss of the cytoplasm indicating that this stage is particularly susceptible to destruction under normal conditions in the host. Most of the individuals seen range from 75 to 125μ in length.

The organs of *Trichomitus* (fig. A, 5) consist of the cytostome (*cyt.*), nucleus (*n.*), food vacuoles (*f. vac.*), and the neuromotor organ system. We will now consider these organs with the cytoplasm in detail.

CYTOPLASM

The cytoplasm of *Trichomitus termitidis* is reasonably labile, finely granular, and somewhat alveolar in structure. This lability may be the cause of such abnormal proportions (such as are seen in plate 4, figure 30) rather than the inheritance of the ancestral parabasal suggested above. The dropping off by plasmotomy of the labile cytoplasm has been observed by us in *Trichomonas augusta* (Kofoid and Swezy, 1915). No contractile vacuole is present but food vacuoles (fig. A, 5, *f. vac.*) are found everywhere within the body except about the nucleus. These contain fragments of cellulose from the digestive tract of the termite or coccoid bodies, possibly bacterial (pl. 3, fig. 5). Defecation of undigested fragments has not been seen.

Upon treatment with neutral red a large number of food particles or metaplasmic droplets stain deeply. They lie scattered throughout the cytoplasm and are larger near the center of the body (fig. A, 8).
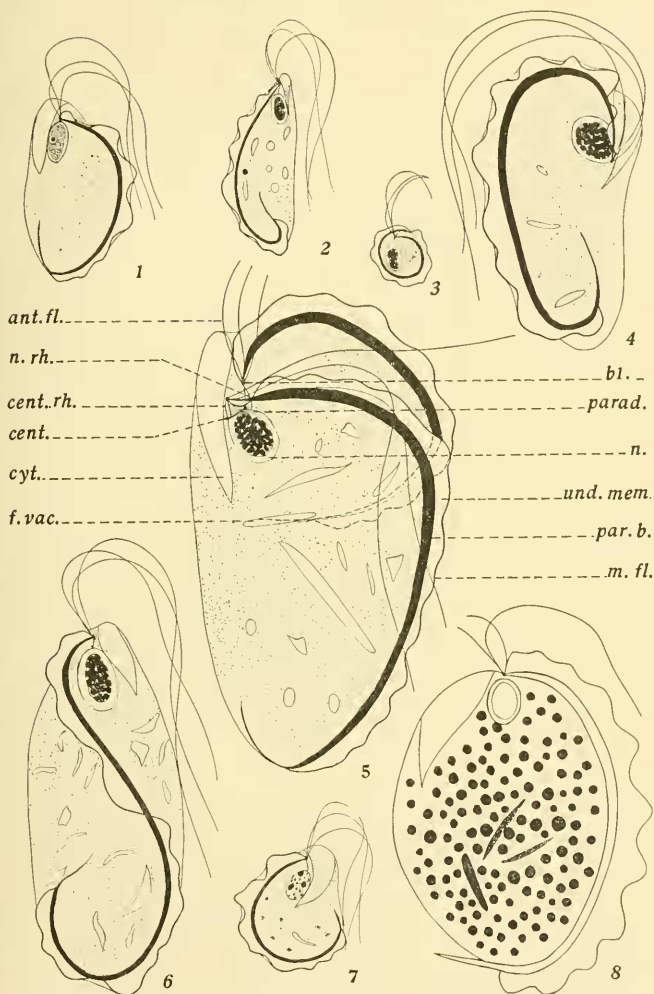
Fig. A. A series showing variations in size in *Trichomitus termitidis*. Figure 8 is drawn from individual stained with neutral red.

Abbreviations: *ant. fl.,* anteror flagella; *bl.,* blepharoplast; *cent.,* centrosome; *cent. rh.,* centrosome-rhizoplast; *cyt.,* cytostome; *f. vac.,* food vacuole; *m. fl.,* marginal flagellum; *n.,* nucleus; *n. rh.,* nuclear-rhizoplast; *par. b.,* parabasal body; *parad.,* paradesmose; *und. mem.,* undulating membrane. × 700.

Cytostome

The cytostome lies on the anterior ventral surface at the extreme anterior end of the body. It is a large elongated asymmetrical pocket, slender pyriform in outline but curved on its right side against the nucleus. Its length is about 0.3 that of the body and its width 0.3 to 0.2 its own length. It leads into the cytoplasm near the center of the body. Its large size, great flexibility and its slight projection anteriorly in a prominent lip all indicate its efficiency as a food-grasping and enveloping organ. We have found it during the later stages of binary fission, in the plasmodium of multiple fission, but not in the encysted condition. We have not been able to determine the exact mode of origin of new cytosomes. Its location immediately adjacent to the blepharoplast and nucleus necessitates a high degree of elasticity, integrity and resistance on the part of these organelles and the rhizoplasts arising from them.

Neuromotor System

The use of the term organ system to designate the complex, structurally integrated apparatus which links together the nucleus and motor organs and plays such a distinctive rôle at mitosis, seems justified by the canons of comparative morphology, unless it be that the dogma of the Cell Theory blights such morphological license. This organ system includes the blepharoplast (fig. A, 5, *bl.*) from which spring directly the three anterior flagella (*ant. fl.*), the attached, posteriorly directed undulating membrane (*und. mem.*) with its marginal flagellum (*m. fl.*), and the deeper lying parabasal body (*par. b.*), and a nuclear rhizoplast (*n. rh.*). The centrosome (*cent.*) lies within the centroblepharoplast, emerging at mitosis with its own independent centrosomal rhizoplasts (*cent. rh.*) joining the ends of the paradesmose (*parad.*) to the parent blepharoplast.

It is noteworthy from the standpoint of comparative cytology that the motor organelles, flagella, and undulating membrane terminate in and originate from the centroblepharoplast. The nucleus never loses its connection, by one or more rhizoplasts, with this structure, which also plays a dominant rôle in the drama of mitosis for at this time there springs from it the centrosome, which later divides, forming the paradesmose and its connecting rhizoplasts. To it also is attached the enormously large parabasal body, a reservoir of chromatoidal sub-

stance. This centroblepharoplast is thus most truly a morphological center intimately associated with the motor organs of this cell and their activities during the vegetative phase. At mitosis it dominates not only the extra-nuclear neuromotor system but also the polarization and subsequent movements of the chromosomes within the nucleus (fig. B, and pl. 3, figs. 5–21) as well. It will be difficult to find in any metazoan cell so continuous and complete a control, so pervasive an influence upon cell activities by the cell organ there known as the centrosome, as we find by the centroblepharoplast in *Trichomitus*.

The *centroblepharoplast*, during the vegetative phase of *Trichomitus* (fig. A, 6), is a minute granule about a micron in diameter anterior to the nucleus and attached to the anterior end of a single nuclear rhizoplast (fig. A, 5, *n. rh.*; fig. A, 2). This rhizoplast is a delicate thread easily overlooked. The centroblepharoplast itself is imbedded in the end of the deeply staining parabasal body, and may likewise readily escape detection.

In view of its later history it seems advisable to designate this granule at this period as the centroblepharoplast, since from it emerges the parent centrosome at mitosis. There is, however, no duplicity of structure evident, and there is no granule at any time at the point where the nuclear rhizoplast passing from the centroblepharoplast (fig. B, 1) and later from the blepharoplast proper (fig. B, 5) meets the nuclear membrane. After the centrosome withdraws from the larger granule (fig. B; pl. 3, figs. 6–21), the latter becomes a blepharoplast in the restricted sense of a basal granule from which the flagella originate, having no other function in mitosis, whereas the centrosome emerging from it divides and its daughters form the paradesmose between them, assume a polar position thereon and move to the nucleus (fig. B; pl. 3, fig. 21).

In our investigations of mitosis in the trichomonads (1915, pl. 2, figs. 21, 23; pl. 3, figs. 24, 29) there appeared to be a separation of the polar centrosome-blepharoplast into two granules, one of which, the centrosome, remained in the polar position on the nucleus, and the other, the blepharoplast, usually with the flagella attached, was removed a short distance therefrom. The conditions which we have found in *Trichomitus* where there is a general, more complete and perfectly distinct separation of these two organelles is thus the full accomplishment of the segregation imperfectly realized in *Trichomonas*.

The *flagella* are four in number, the three undifferentiated, equal, anterior ones (fig. A, 5, *ant. fl.*) and the attached posteriorly directed

one included within the undulating membrane as its marginal fiber
(*m. fl.*) and carried out beyond the projecting tip of the parabasal
body as a bit of free flagellum. The anterior flagella usually exceed
the body in length. The posteriorly directed location in our figures
is merely for spatial accommodation, an anterior direction being usual
in life.

The *undulating membrane* (fig. A, 5, *und. mem.*) is attached to the
left side of the body (fig. A, 1) in a sweeping C- or S-shaped curve
reaching to the posterior end of the body (fig. A, 4, 6; pl. 3, figs. 1, 14).
It exceeds the length of the body two to three times in some small
schizonts (pl. 4, fig. 30). The coiling into the S-shaped forms appears
to be an accommodation of the somewhat rigid but elastic parabasal,
when longer than the body, to its location within the cytoplasm.
The membrane always follows the course of the parabasal and remains
adherent to it upon cytolysis (pl. 3, fig. 11). In one case (pl. 3, fig. 4)
a detached membrane consisting only of the marginal flagellum and
the fold of the protoplasmic pellicle running from the parabasal around
the flagellum, was found free in a smear preparation. The membrane
and flagellum are thrown into twelve to twenty subequal, subequi-
distant waves of contraction which fade out in the distalmost end.

The parabasal body (fig. A, 5, *par. b.*) is a rigid, elastic, deeply
staining, chromatoidal rod lying at the base of the undulating mem-
brane in the peripheral plasma of the body. Its C- or S-shaped course
appears to determine the direction of that membrane. Its length
usually exceeds that of the body by 10 to 25% and its diameter,
2 to 3.5μ, is greatest somewhat anterior to its middle. From this
region it tapers gradually toward either end, terminating anteriorly
at the centroblepharoplast (fig. A, 6), or in mitosis at the blepharo-
plast proper (fig. A, 5), and posteriorly at its junction with the
marginal flagellum which projects beyond their union for a short dis-
tance as a free lash. It stains densely with haematoxylin and con-
stitutes the dominating feature of the organism in all preparations
and in life. It shows in stained sections (pl. 3, figs. 3 and 3a) a
differentiated structure consisting of an outer deeply staining shell
less than a fifth of its diameter in thickness and a less deeply stained
core. This core is traversed by wedgelike discs of the cortical sub-
stance, which arise principally on the concave face and fade away
towards the opposite side.

We have elsewhere (Kofoid and Swezy, 1915; Kofoid, 1917) inter-
preted the parabasal body as a reservoir of substances utilized by the
neuromotor system in motor activities. It is obvious on observation

that locomotion by *Trichomitus* in the midst of the seething mass of parasites in the digestive tract of *Termopsis,* involves not a little expenditure of energy. It is also conceivable that the conditions of life therein are subject to marked variations incident not only to the food and feeding of the host but also to the varying constituents of the enormous mass of parasitic associates and their changing metabolism due to phases of their reproductive activity. Biochemical changes of no small import are consequently a feature of this creature's environment. That some of these are peculiarly fatal to *Trichomitus* is evident from the unusual numbers of moribund or cytolized individuals, each represented by a more or less decadent nucleus and its attached neuromotor apparatus, which may be found in most smear preparations.

Considerable changes in extent and volume of this structure are apparent upon an inspection of our figures, and even more so in our preparations. These are indicative of changes resulting from metabolism, or multiple fission, or both. In addition to the storage or reservoir function it is apparent that the parabasal in *Trichomitus* serves also as a somewhat rigid organ of attachment for the undulating membrane.

The other organs of the neuromotor system, the paradesmose and its rhizoplasts will be discussed in connection with mitosis. The paradesmose is a more or less temporary organ in most trichomonads, but in *Trichomitus* the organism appears to pass a much greater part of its existence in what is comparable to the prophase stage of trichomonad mitosis, so that the paradesmose is actually present, suspended by rhizoplasts from the blepharoplasts (fig. A, 5), and the whole neuromotor system is in some phase of duplication in many of the individuals which we have seen. The stage with a single centroblepharoplast and rhizoplast (fig. A, 1, 2, 6) is relatively much less common in this species than in other trichomonads. This prolongation of the prophase is the first step towards diplozoic organization such as we find realized in *Giardia*, where nuclear division is added to that of the duplication of the neuromotor system with the resulting formation of a coördinating system for multicellular organization.

Nucleus

The nucleus (fig. A, *n.*) is a symmetrical ellipsoidal structure, or even ovoidal or pyriform, with the wider end posterior (fig. B, 1). The longer axis is two to three times the shorter one in length. It

lies in or near and parallel to the major axis of the body on the left
side of the cytostome within a short distance of the anterior end of the
body. It shows distinctly a peripheral clear zone which is somewhat
regularly chambered (fig. A, 1, 4, 5; pl. 3, fig. 9) as we have found it
also in *Trichonympha*. This zone surrounds the dark, dense and often
seemingly undifferentiated central chromatin mass. On heavy destain-
ing this central mass is at times resolved into fairly uniform rounded
granules (pl. 3, fig. 2) which appear to have some special relation
to the elements of the chambered zone, indicating the possibility of a
persistent organization of the nucleus. In some instances resting
nuclei (fig. B, 1; pl. 3, fig. 5) show large deeply staining granules
resembling nucleoli but these are as a rule absent. It is quite possible
that these may be end knobs of emerging chromosomes.

The size of the nucleus ranges from 10 to 20μ in length whereas
that of the body ranges from 16 to 200μ or even more. Although
larger individuals have larger nuclei the increase in the volume of the
cytoplasm is many-fold greater than is the increase of the nucleus in
these giant forms.

One of the most significant and striking features of the life his-
tory of *Trichomitus* in the digestive tract of *Termopsis* is the repeated
and seemingly constant occurrence of large numbers of isolated neu-
romotor systems with the nucleus attached but no enveloping cyto-
plasm. It is evident that the delicate pellicle is easily destroyed, and
the labile cytoplasm escapes. Such an isolated structure in late pro-
phase with duplicated neuromotor systems but degenerate nucleus is
seen in plate 3, figure 11. These occurrences afford indisputable
evidence of the organic continuity and structural integration of the
neuromotor system of *Trichomitus* and of its direct and efficient
physical connection with the nucleus. One of us (Swezy, 1915a) has
noted a similar phenomenon in *Hexamitus*, a diplozooic polymastigote.

Still more significant is the fact that such isolated systems are
still capable of flagellar activity and locomotion after the destructive
process of cytolysis of the cytoplasm. They continue to move for some
time in smears of the contents of the digestive tract mounted in tap
water. The very large number of isolated systems found in some
smears is indicative of a considerable period of persistence of the
isolated neuromotor system and nucleus after the loss of cytoplasm.
It is obvious that grave limitations on such activities must arise as a
result of the loss of the cytoplasm. The nucleo-cytoplasmic reactions
are suspended, nutrition is impeded if not wholly suspended, and
rapid exhaustion is accelerated by the loss.

MITOSIS

Owing to this prolongation of the prophase of mitosis in *Trichomitus termitidis* an exceptional opportunity is afforded for a detailed study of the behavior of the neuromotor system during mitosis. This is made possible by reason of the fact that the centroblepharoplast is the center of the neuromotor system and the point of origin of structures and processes playing the main rôle in mitosis.

The phases of mitosis recognizable in the division of *Trichomitus* are those of the metazoan cell, but, as shown in our (1915) discussion on mitosis in trichomonads, considerably differentiated by the association of the extra-nuclear organelles of the cell in the protozoan from that in the usually simpler metazoan unit. In *Trichomitus* these differences, in consequence of the massive development of the neuromotor system, are even more developed than in the other trichomonads. They consist mainly in the sharp separation of centrosome and blepharoplast and the excessive prolongation of the prophase.

The *resting stage* of *Trichomitus* has a single nuclear rhizoplast running from the centroblepharoplast to the anterior end of the nucleus where it is attached to the membrane without evidence of enlargement into a centrosome on the nuclear membrane. This rhizoplast is often very short (fig. A, 1) and is never very long. The nucleus has in this period a dense, coarsely and uniformly granular central chromatin mass in which no polarization or evidence of chromosome formation is present.

The *prophase* (fig. B; pl. 3, figs. 1, 2, 5–15) is a prolonged one and in it the duplication of the entire neuromotor system takes place by division and outgrowth from the centroblepharoplast. The initial step is the splitting of the centroblepharoplast and the nuclear rhizoplast (fig. B, 1). It appears in some cases (pl. 3, fig. 1) that the rhizoplast may divide at the nuclear membrane first and split distally towards the centroblepharoplast as though these were directly upon the nuclear membrane at the anterior end of the nucleus. At the same time the centroblepharoplast separates into its constituent centrosome and blepharoplast, with the latter immediately dividing, one granule taking a single flagellum, the new parabasal body and one of the two nuclear rhizoplasts (fig. B, 1). The granule remaining attached to the old parabasal body takes the remainder of the flagella and the second nuclear rhizoplast. As these two blepharoplasts separate a thread is drawn out from each attaching them to the centro-

some (fig. B, 1). This is followed by a division of the centrosome.
As the two new centrosomes move apart a darkly staining line or bar
is drawn out between them, the paradesmose (fig. A, 5, *parad.*; fig. B).

The rhizoplasts connecting the centrosomes with the blepharo-
plasts gradually elongate with the paradesmose also increasing in both
length and thickness. The latter structure with its connected centro-
somes moves down until it comes to rest upon the nuclear membrane
(fig. B). As a result of this the centrosome-rhizoplasts come to have
the same length as the nuclear rhizoplasts. All four of these rhizo-
plasts are exceedingly delicate, particularly the nuclear ones and the
latter especially long escaped our notice. Since the two are rather close
together (fig. B, 5) their distinctiveness may be easily overlooked.
The centrosomes (fig. A, 5, *cent.*) are minute knobs on the ends of the
paradesmose, which is a stout, heavy, sometimes granular, deeply
staining bar. They are not always visible as expansions of the bar and
are never seen detached from it.

In the meantime the new undulating membrane and parabasal
body have reached sizes equal to the ancestral ones (pl. 3, fig. 11;
fig. B, 2–6). The parabasal body first appears as a slender dark
thread growing posteriorly subparallel to the old parabasal in the
peripheral cytoplasm (pl. 3, fig. 5). With the beginning of the
formation of the paradesmose (fig. B, 2) the new flagella have all
formed by outgrowth from the daughter blepharoplasts, two anterior
ones and a posteriorly directed one as a marginal filament from the
blepharoplast attached to the new parabasal, and only one anterior
one from that attached to the old parabasal. At first (fig. B, 2) the
new undulating membrane is very narrow but it soon attains full
structural size and functional efficiency (pl. 3, fig. 11). With the
completion of these structures by growth the duplication of the neuro-
motor system is accomplished.

Up to this time the changes visible within the nuclear membrane
have been very slight. The chromatin granules or chromomeres grow
larger and darker, and evidences of polarization appear in the linear
grouping of the granules (fig. B, 6; pl. 3, figs. 6–10) which cul-
minates in the emergence of linear V-shaped chromosomes. During
this process a deeply staining cone-shaped extension of the central
chromatin mass projects anteriorly until it comes in contact with
the paradesmose (fig. B, 6; pl. 3, figs. 6–8). As this disappears the
V-shaped chromosomes become more evident in the central mass, and

gradually spread out below the paradesmose as though hung across
a string suspended from its ends (pl. 3, figs. 10, 12, 13).

The number of chromosomes is rather obscure since the loops are
at all times rather closely entangled. It appears to be twelve or
thereabouts. In the earlier phases each is composed (pl. 3, fig. 10)
of a line of distinct granules, like chromomeres, but these fade out as
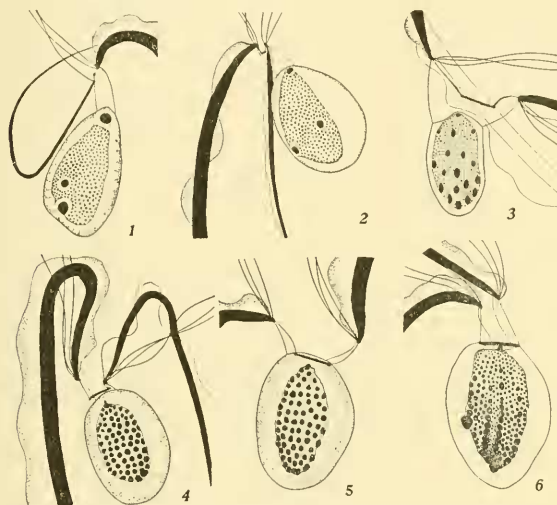the metaphase approaches.

Fig. B. Development of paradesmose in *Trichomitus termitidis.* 1. Out-
growth of new parabasal body after separation of centrosome and blepharoplast
and division of latter. 2. Centrosome divided and paradesmose forming be-
tween them; new flagella and undulating membrane formed. 3. Elongation of
paradesmose as it moves down to the nucleus with the lengthening of the
centrosome-rhizoplasts. 4. Later stage of same, condensing chromatin in nucleus
with cone-shaped projection starting towards the nuclear membrane and parades-
mose. 5. Paradesmose attached to nuclear membrane. 6. Attachment of central
chromatin mass to paradesmose and formation of spindle; beginning of formation
of chromosomes.   × 1575.

The *metaphase* is obscured by the fact that there appears to be
no arrangement of the chromosomes in an equatorial plate and no
amphiaster phase of the nucleus. The nuclear structures appear to
conform their arrangement to the stout bar-shaped paradesmose and
not the latter mould itself to the configuration of the nucleus as in
Trichomonas and *Eutrichomastix* (Kofoid and Swezy, 1915).

The loops or V-shaped chromosomes are gradually drawn by the angle of the V towards the two ends of the paradesmose (pl. 3, figs. 15, 17, 18). It is possible that each original loop is split length-wise during this movement but the evidence for it is by no means clear. During this process the loops shorten, thicken, and stain more deeply, so that when they have finally parted (pl. 3, figs. 16, 20, 21) they form chrysanthemum-shaped rosettes.

The *anaphase* is brief and is also dominated by the stout parades-mose, which continues to produce a one-sided, asymmetrical grouping of the two groups of parting chromosomes and to modify the constric-tion of the nuclear membrane so that it is also one-sided. The nuclear membrane remains intact throughout the whole process of mitosis. In the late anaphase constriction is completed, the nuclei separate and move apart (pl. 4, fig. 22), stretching out the paradesmose be-tween them as a result of the uncoördinated activities of the two daughter neuromotor systems, which are attached to the nuclei by their rhizoplasts.

The *telophase* (pl. 4, fig. 36) ensues before plasmotomy. In it the chromatin of the massed chromosomes rounds up in the central mass and the clear zone reappears and the ellipsoidal form is resumed. The paradesmose also fades away, and the centrosome merges with the daughter blepharoplast which, by the shortening of the nuclear rhizoplast, comes to lie closer to the nuclear membrane, thus bringing the schizont back to the nuclear condition prior to mitosis.

The process of mitosis in *Trichomitus* is similar to that in other trichomonads in that the nuclear membrane remains intact through-out the process, the extra-nuclear paradesmose arises between the daughter centrosomes, and the duplication of the neuromotor system proceeds from the centroblepharoplast and takes place prior to divi-sion of the chromosomes. It differs in having a distinct separation of centrosome and blepharoplast for a long period, in having a rigid bar-shaped paradesmose, and in the large size and greater elongation of the chromosomes. This higher specialization is conditioned by crowded conditions of parasitic life in association with other parasites of relatively complex organization. The conditions of locomotion in this association and the excessive amount of stimulation consequent thereon are causes conducive to the extraordinary development of the neuromotor apparatus in *Trichomitus* and the resulting modifications in mitosis.

BINARY AND MULTIPLE FISSION

Both of these processes take place frequently in *Trichomitus*. There is much evidence of a high death rate in this species within the digestive tract of its host. This is compensated for by rapid multiplication. For this also there is abundant evidence in our material.

The distinction between stages of binary and multiple fission is not readily made in all cases in early stages. The earliest phases of both are obviously the same in mitotic phenomenon. Binucleate plasmodia may lead on to further division when prophases appear in their nuclei (pl. 4, fig. 23). When, however, no later prophase phenomena are evident and the organism is not unusually large (pl. 4, fig. 36) binary fission only may be expected. It also occurs in the small cysts (pl. 4, fig. 26) and small free forms (pl. 4, fig. 29).

Multiple fission, on the other hand, occurs in large individuals and leads to the formation of, presumably, eight-celled plasmodia.

One such large plasmodium with six constituent zooids is seen in plate 4, fig. 28. It is possibly in plasmotomy and has lost two of its members. Not all multiple fission plasmodia are as large as this. Smaller ones, in which the first division has been completed and the second initiated, are frequently found (pl. 4, figs. 23, 25, 35).

The process is one of three repeated divisions prior to plasmotomy with the formation of an eight-nucleate somatella and its subsequent disintegration into its constituent zooids by their detachment singly or in groups. These somatella or plasmodial stages are exceedingly mobile and the constituent individuals shift about without seeming order of arrangement. The uncoördinated movements of the powerful neuromotor apparatus of the individual zooids finally result in their separation. The connecting paradesmoses are lost long before this separation.

No trace of unequivocal sexual phenomena has been detected. Large and small individuals simulating macrogametes and microgametes and gametocytes are present. Binucleate individuals without evidence of recent division occur (pl. 4, fig. 36), simulating zygotes, and similar associations are found in cysts (pl. 4, fig. 26). There is, however, no evidence of maturation divisions leading to gamete formation, no sexual behavior detected, and no evidence of the fusion of gametic nuclei. In the absence of such evidence any conclusions as to the possibility of sexual reproduction in this organism must be held in abeyance.

ENCYSTMENT

Associated with the vegetative and fission stages of *Trichomitus* in a few hosts but not all, we have found many small individuals (pl. 4, figs. 24, 29–33) in which binary fission is occurring and in which there is a tendency for the body to round up into a spheroidal or ellipsoidal mass. These small sizes may result from rapid fission without compensating growth or from plasmotomy of part of the cyto- plasm. In these same hosts occur also numerous ellipsoidal cysts about 13 by 20μ with a deeply stained network with thickenings at the nodes spread over the surface (pl. 4, figs. 26, 27). The cyst wall is double and the network is due to the accumulations of some stainable substance between the walls. Within the cyst is a single individual (pl. 4, fig. 31) with a very long parabasal and undulating membrane making nearly two complete coils, such as might result from the coiling up of an individual with abnormally large neuromotor system (pl. 4, fig. 30). In other cases two such individuals (pl. 4, fig. 26) are found within the cyst. This might result from encystment after or during mitosis but prior to plasmotomy. Such cysts may facilitate the carry- ing over of infection from one individual host to another. They have all the indications of being resistant stages.

RELATIONSHIPS

This is a species of *Trichomitus*, a genus founded by one of us (Swezy, 1915*b*) for the reception of a minute and simple trichomonad from amphibians. In its vegetative phases the form here described has the morphological features of *Trichomitus parvus* Swezy, namely, three anterior flagella, undulating membrane, parabasal, and no axo- style. It differs greatly in size, in the massive development of the parabasal, and at mitosis in the distinct separation of centrosome and blepharoplast. Binary and multiple fission were followed in the species from amphibians but no trace of such separation was detected.

Such a difference as this might justify generic separation but it might be impracticable to apply it in future diagnosis of any species of the genus which may come to light. The difference is, however, of such morphological import as to justify subgeneric separation. We accordingly assign it to

Trichomitopsis subgen. nov.

Trichomitus with centrosome separated from blepharoplast at mitosis. Type species *Trichomitus termitidis* sp. nov. from *Termopsis angusticollis* Walker.

SUMMARY

1. *Trichomitus termitidis* sp. nov. occurs in the intestinal tract of *Termopsis angusticollis*. It is apparently not pathological to its host, is never attached to the wall and feeds on the débris of the intestinal contents.

2. It has a highly developed neuromotor system with parabasal body, undulating membrane, centroblepharoplast and flagella attached by a rhizoplast to the nucleus.

3. Binary fission occurs frequently. Mitosis is marked by the development of a large paradesmose following the separation of the centrosome from the blepharoplast. One schizont retains the old parabasal body and membrane, while new ones are formed for the other.

4. Multiple fission results in the formation of an eight-zooid somatella followed by plasmotomy.

5. Owing to the great differences in the process of mitosis between *Trichomitus parvus* and the new species, *T. termitidis*, subgeneric distinction is given to the latter, as we assign it to the new subgenus *Trichomitopsis*.

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EXPLANATION OF PLATES

All figures of *Trichomitus termitidis* sp. nov., from material stained with iron alum haematoxylin. Magnification 625, unless otherwise stated.

PLATE 3

Fig. 1. Trophozoite in early prophase of division with nuclear rhizoplast divided.

Fig. 2. Isolated neuromotor apparatus of the same stage with nucleus attached.

Fig. 3. Sagittal section of the parabasal showing the outer deeply staining shell and the inner core. × 1250.

Fig. 3a. Cross-section of the same. × 1250.

Fig. 4. Isolated undulating membrane with darkly staining marginal flagellum. × 1250.

Fig. 5. Early prophase with outgrowing of new parabasal body.

Fig. 6–10. Early prophase stages showing gradual condensation of chromatin into definite chromosomes. New parabasals, undulating membranes and flagella complete.

Fig. 11. Isolated neuromotor system in prophase stage of division.

Figs. 12, 13. Spindle forming with chromosomes attached to it by the angle of the V. × 1250.

Fig. 14. Prophase. Note relative lengths of parabasal bodies.

Figs. 15, 17. Separation of chromosomes into two groups. × 1250.

Figs. 16, 18–20. Anaphase with paradesmose elongating. × 1250.

Fig. 21. Beginning of constriction of the nuclear membrane. × 1250.